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(54) 3-indolepyruvic acid derivatives their method of production and therapeutic use

3-Indolbrenztraubensäure, Verfahren zu deren Herstellung und deren therapeutische Verwendung

Dérivés de l'acide 3-indol-pyruvique, méthode pour leur préparation et leur utilisation thérapeutique

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- **SYNTHESIS, no. 7, July 1988, pages 514-517, Stuttgart, DE; G. TARZIA et al.: "Alkyl 2-(diphenylmethyleneamino)acrylates in the synthesis of alpha-amino acids"**

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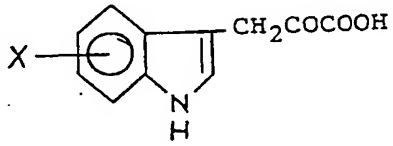
Description

The present invention refers to a process for the production of 3-indolepyruvic acid derivatives, substituted on the benzene moiety.

The compounds according to the present invention are characterized by the presence of substituents on the benzene ring of indole and are represented by the general formula I



(I)

in which X is methyl, methoxy, hydroxyl or halogen.

Reference to the prior art

3-indolepyruvic acid has shown itself to possess extraordinary biological and pharmacological activity because, having double bonds conjugated into enole configuration, it is capable of capturing oxygen free radicals (especially .OH) and therefore of transforming itself into kynurenic acid, which in international literature appears as the most important physiological antagonist of excitatory aminoacids. Said aminoacids are substances capable of irreversibly destroying neurons in degenerative diseases, such as cerebral ischemia, ageing, epilepsy, etc. (see for example Lancet, II, 140-143, 1985; Neurosience Letters 48, 273-278, 1984).

The properties of 3-indolepyruvic acid as a capturer of free radicals have been described in the published European application No. 0362152. The capability of 3-indolepyruvic acid to transform itself into kynurenic acid has, on the other hand, been described in the international patent application WO 88/09789. In the latter application have also been described several derivates of 3-indolepyruvic acid, obtained by esterification or amidation of the carboxylic acid.

Summary of the invention

It has now been found, that 3-indolepyruvic acid derivatives obtained by substitutions on the benzene ring of indole, are compounds of even greater interest, as they can antagonize the harmful effects of oxygen free radicals and of excitatory aminoacids in a manner even more efficacious than 3-indolepyruvic acid and its derivatives described in the prior art.

Object of the present invention is a process of production of said derivatives represented by formula (I), which consists in starting from benzene, firstly performing the substitutions on the ring, then transfroming the substituted benzene into substituted indole, then transforming the latter into substituted tryptophan, and finally operating on the latter to obtain 3-indolepyruvic acid derivates.

Synthesis of the compounds of formula (I)

The direct insertion of a substituent into the benzenoid structure of indolepyruvic acids is known to be prohibited by the characteristics of intrinsec instability of the molecule (see for example Chemical Review 83, 321, 1987). For similar reasons the direct substitution on the tryptophan molecule is also impossible.

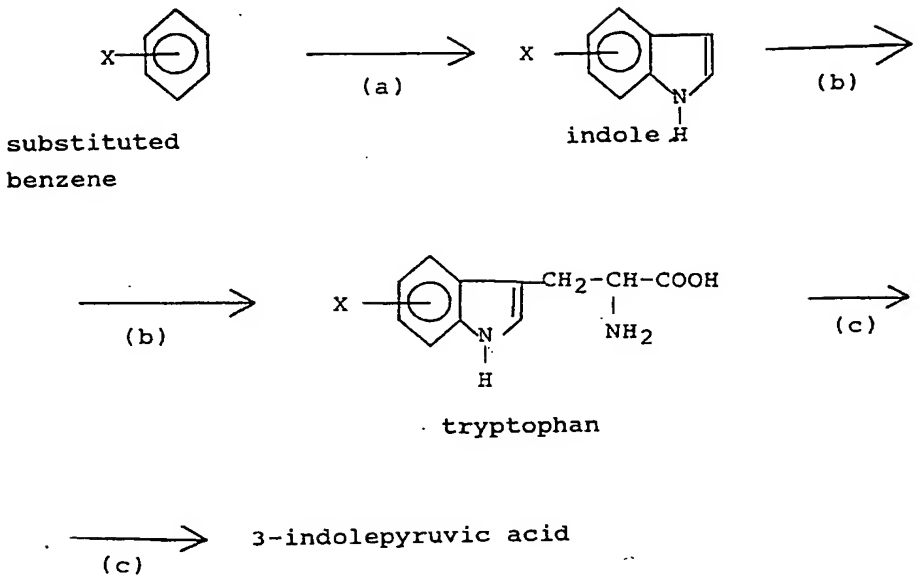
For the preparation of benzenoid substituted indolepyruvic acids, according to the present invention, the method has been used of starting from substituted benzene, transforming it into substituted indole, then passing to the corresponding substituted tryptophan and finally preparing the substitutes indolepyruvic acids.

The synthesis of the indole ring is well documented by a rich chemical literature (see for example "Synthesis of the indole nucleus" in : Indoles, part I, J, Wiley & Sons, 1972; "The Chemistry of indoles", Chapter III, Academic Press, 1970; "Contemporary heterocyclic chemistry", Chapter V, J. Wiley and Sons, 1982; etc.).

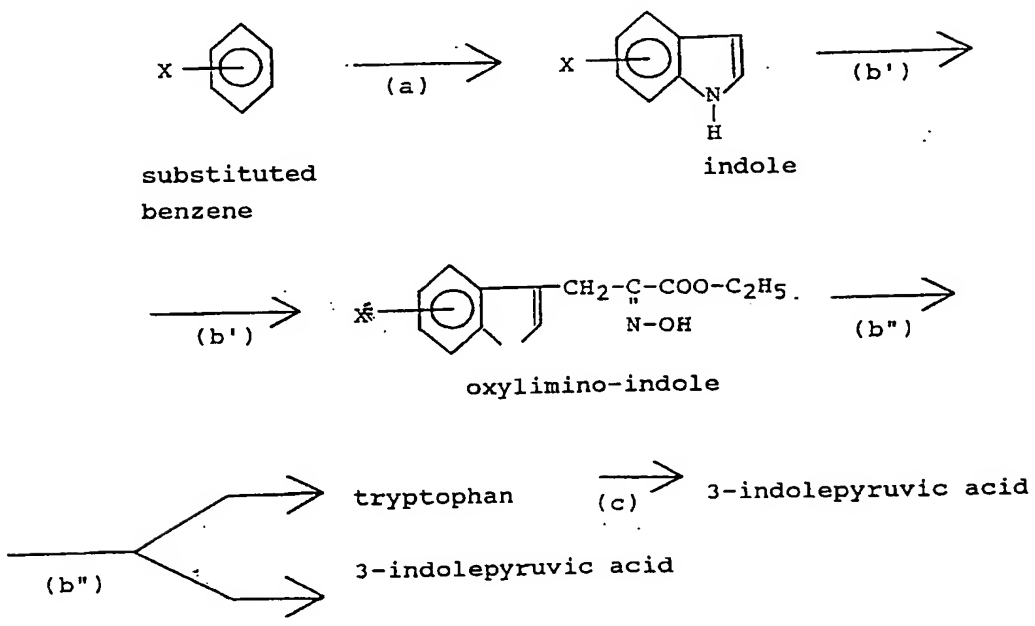
In general all the synthetic approaches that want positions 2 and 3 of the indole ring free or easily freed, start from suitably substituted benzene and differ from one another only in the sequence of reactions which bring about the formation of the hetero-aromatic (pyrrolic) part of the indole ring.

The transformation of the indole ring to tryptophan is also supported by a large amount of literature. Said transformation can be brought down to two general methods, which are resumed in the following table:

First method of synthesis (prior art)



Second method of synthesis (according to the invention):



The first method of synthesis shows the intermediate transformation of the indole into the corresponding Mannich base, that is gramine, thus taking advantage of the capacity of the Mannich base to condense, in an alkaline catalitic environment, with derivates of a malonic type (alkylation of compounds with active methylene).

See for example "The Chemistry of the amino acids" chapter 39, J. Wiley and Sons, 1961; "Chemistry and biochemistry of the amino acids" chapter 7, Chapman & Hall, 1985.

The second method of synthesis according to the present invention on the contrary shows the direct alkylization of the indole ring, preformed with a suitable structure containing the amino acid part of tryptophan in a masked form (see for example J. Organic Chemistry 49, 540, 2657 and 4409, 1984; Tetrahedron 38, 2051, 1982). Said second method furthermore permits the direct transformation of the intermediate oxylimine-indole into the corresponding indolepyruvic acid derivate, along with the passage through the corresponding tryptophan.

For the transformation of the substituted tryptophan on the benzene ring into the corresponding 3-indolepyruvic acid derivate various methods can be used. A particularly advantageous method is described in European patent publication No. 0227787.

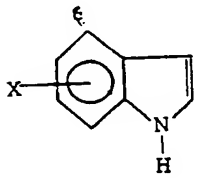
Examples of preparation

Preparation of indole derivates

Where possible the substituted indole derivates on the benzene ring have been acquired from commercial sources. For example the 4- and 6-chloroindole, 5-bromoindole, 4-, 5- and 7-methylindole, 4- and 5-hydroxylindole, 5- and 6-fluoroindole have been obtained from Sigma Chemical. 5-chloroindole from Janssen Chimica; 6-methylindole from Aldrich, etc.

Otherwise they can be obtained by the classical methods previously cited, starting from the corresponding substituted benzene.

In this way general compounds have been obtained of formula II

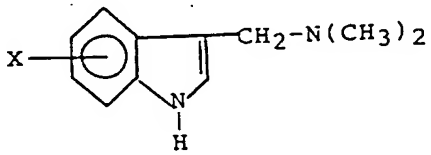


II

in which X has the meaning indicated above.

Preparation of gramine derivates

The substituted indole derivates of formula (II) were made to react with dimethylamine and aqueous formaldehyde to form general compounds of formula III



III

in which X has the meaning indicated above.

Example 1 - Synthesis of the compound 6-chloro-3(dimethylaminomethyl)-indole, or 6-chloro-gramine

Glacial acetic acid (0,4 ml) was added dropwise,to an aqueous solution at 33% (w/v) of dimethylamine (0,4 ml), cooled in an ice bath, at a speed such as never to exceed the temperature of 5°C. Under continuous stirring and in an ice bath, were added in succession an aqueous solution of formaldehyde (0,2 ml at 40% w/v) and then 420 mg of 6-chloroindole, which in approximately 10 minutes dissolved in the reaction mixture with the development of heat. The reaction mixture was left at room temperature for approximately 16 hours. The solution was then poured into NaOH 2N (10 ml) and extracted with ethyl ether (3x15 ml). The organic phase was washed with saturated NaCl/H_2O (2x10 ml)

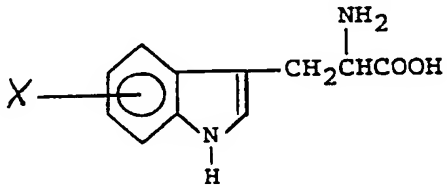
and dried on anhydrous magnesium sulphate overnight. On evaporation of the solvent, a residue was obtained, of practically pure 6-chloro-gramine, equal to 550 mg (yield 95%).

Example 2 - Preparation of 6-chloro-tryptophan

Under inert atmosphere and rapid stirring 52 mg of NaOH are pulverized into anhydrous xylene (20 ml), heating the suspension at approximately 90°C. 550 mg of 6-chlorogramine (prepared as described in example 1) and 460 mg of ethyl-acetamide-cyanoacetate are then added. The mixture is refluxed for approximately 7 hours and the development of dimethylamine, extremely vigorous at the start of the heating, practically ceases after about 6 hours of heating. The reaction mixture is cooled for 12 hours at 5°C and then filtered to recover the abundant precipitate. This crude solid is treated with a hot solution of benzene/absolute ethanol, and the insoluble material is hot-filtered. The solution is left to rest at 5°C for 12 hours. Crystals slowly separate, which are filtered and dried. 750 mg of ethyl-a-acetamine-(6-chloro-indol-3-il)-propionate are recovered, with a yield of 84%. 600 mg of this compound are added to 15 ml of water, containing 40 mg of NaOH and the mixture is boiled for 24 hours. During this time the solid material dissolves and ammonia is developped. After the reflux time, the reaction mixture is cooled at room temperature and neutralized with acetic acid at 50%. It is filtered, recovering and washing with cold water the solid which has formed. After drying 410 mg of 6-chloro-DL-tryptophan are recovered, with a yield of 95%.

Example 3

In a similar way are prepared the tryptophan derivates of general formula IV



IV

in which X has the meaning indicated above.

Example 4 - Preparation of 6-chloro-indol-3-il-pyruvic acid

To 600 mg of 6-chloro-DL-tryptophan, suspended in 5 ml of methanol, are added, at room temperature, 0,230 ml of triethylamine and the mixture is stirred for approximately 10 minutes. Then, rapidly and under stirring, pyridine-4-carboxyaldehyde (0,240 ml) is added. After approximately 5 minutes, total dissolution of the suspension is obtained. Stirring is continued for 10 minutes. 122 mg of anhydrous ZnCl2 are then added, stirred for 10 minutes, followed by 0,490 ml of 1,8-diazabicyclo-(5,4,0)-undec-7-ene. The solution becomes orange-red in colour. It is left under stirring for approximately 80-90 minutes at room temperature. The limpid red solution is then quickly added dropwise and under rapid stirring to 100 ml of HCl 1N preheated to 50°C. After approximately 10 minutes from completion of the addition a spontaneous precipitation occurs, increasing in time, of a yellowish solid. The mixture is left for a further 20 minutes at 55°C, then for approximately 3 hours it is cooled to room temperature. The abundant precipitate is filtered, washed with acidic water and dried. 195 mg are recovered, with a yield of 54%.

Example 5

In a similar manner are prepared compounds of the general formula I, among which are indicated as examples:

4-chloro-indole-3-yl-pyruvic,
6-chloro-indole-3-yl-pyruvic,
5-bromo-indole-3-yl-pyruvic,
4-methyl-indole-3-yl-pyruvic,
5-methyl-indole-3-yl-pyruvic,
7-methyl-indole-3-yl-pyruvic,
4-hydroxy-indole-3-yl-pyruvic,
5-hydroxy-indole-3-yl-pyruvic,
5-fluoro-indole-3-yl-pyruvic,
6-fluoro-indole-3-yl-pyruvic,

5-chloro-indole-3-yl-pyruvic,
6-methyl-indole-3-yl-pyruvic.

Synthesis through indole hydroxylimine intermediates (second method of synthesis according to the invention)

Derivates of general formula I can also be obtained by means of the second method of synthesis mentioned herein above. This comprises a first condensation reaction, in a catalytic environment basic for anhydrous Na_2CO_3, between the substituted indole and the oxime of the ethyl-3-bromopyruvate ester (prepared in situ). From this intermediate are easily obtained both the corresponding substituted tryptophan (by reduction with $TiCl_3$) and the derivate of indolepyruvic acid, by means of the hydrolitic hydrogenization with sodium hypophosphite, in the presence of Nickel-Raney catalysts in a buffered environment.

Example 6 - Synthesis of 3-indolepyruvic acid

By means of the second method of synthesis, it has also been possible to perfect a new and advantageous method for the synthesis of 3-indolepyruvic acid without substitutions, through the condensation reaction of the unsubstituted indole and ethyl-3-bromopyruvate ester oxime prepared in situ, and thus the hydrolytic hydrogenization of the intermediate compound with sodium hypophosphite in the presence of Nichel-Raney in a buffered environment.

The preparation is described herebelow.

840 mg of ethyl-3-bromopyruvate ester oxime were mixed with 940 mg of indole and 680 mg of anhydrous Na_2CO_3 in absolute dichloromethane. The oxime was prepared quantitatively according to the method of Ottenherjm, Tetrahedron Letters 5143, 1978. The mixture was then stirred at room temperature for approximately 12 hours. After having removed the solvent at reduced pressure, the residue was taken up with ethyl acetate and the whole was washed with water. Drying was performed on anhydrous magnesium sulphate. After having eliminated the drying means, concentration was carried out at low pressure and filtering was performed through silica with a mixture of petroleum ether (30/50)/ethyl acetate (1:1). After having eliminated by crystalization the indole, cromatography was performed on silica, recovering 869 mg of the oxyliminic derivate of ethyl-3-(indole-3-yl)-2-hydroxylimino propionate, with a yield of 80%. 490 mg of this compound were dissolved in 20 ml of ethanol/acetate buffer pH 5,0 (2:1). Under stirring was added an aqueous suspension of sodium hypophosphite (880 mg) containing Nichel-Raney (10 mg). This was brought under stirring to the temperature of approximately 50 °C for 3 hours. The reaction mixture was then filtered on celite and the solution was rapidly added dropwise and under energetic stirring to a large excess of HCl 1N, preheated to 55°C (100 ml). After approximately 5 minutes from addition, a considerable precipitation of a canary-yellow colour was seen, which increased with time. Heating was continued for a further 15 minutes and the whole was then allowed to return to room temperature. After 3 hours the precipitate was filtered and then washed with acidic water. After drying 330 mg of 3-indolepyruvic acid were recovered, with a yield of 80%.

6-OMe-IPA	1x10-4 M	200 mg/Kg ip	100 mg/Kg ip
6-Cl-TRP	1x10-3 M	1000 mg/Kg ip	200 mg/Kg ip
IPA = 3-indolepyruvic acid; TRP = Tryptophan; IC50 = dose of compound necessary to reduce by 50% the harmful effect to be added to the in vitro tests or to be administered to mice.			

In these pharmacological tests, in vitro and in vivo, the compounds synthetized have shown themselves to possess characteristics for their development as inhibitors of the damage caused by free radicals and by excitatory amino acids.

It is equally evident that simple salts and esters of the above mentioned compounds have a similar behaviour.

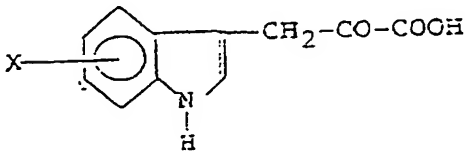
The compounds can be pharmacologically employed in situations such as epilepsy, cerebral ischemia, ictus, Alzheimer's disease, cerebral deficiency of kynurenic acid.

The administration can be made by means of pharmacological compounds containing the active substance in a dose from 2 to 20 mg/Kg body weight in a "per os" or rectal administration, and in a dose from 1 to 10 mg/Kg body weight in a parenteral administration.

For oral, parenteral or rectal administration, the usual pharmaceutical forms can be used, such as pills, capsules, solutions, suspensions, injections, suppositories, in association with pharmaceutically acceptable vehicles or diluents and excipients.

Claims

1. A process for the production of 3-indole pyruvic acid derivatives substituted on the benzene moiety, represented by formula I



I

in which X is hydrogen, halogen, methyl, methoxy or hydroxil, starting from a nucleus substituted indole, characterised in that:

said substituted indole is condensed, in a basic catalytic environment, with 3-bromo-pyruvic acid lower alkyl ester oxime to obtain lower alkyl-3-(substituted-indole-3-yl)-2-hydroxylimine propionate; and said oxylimine intermediate compound is transformed into the corresponding substituted tryptophan by reduction with aqueous $TiCl_3$, and

said substituted tryptophan is transformed into said substituted 3-indole pyruvic acid in a per se known manner.

2. A process according to claim 1, in which said oxylimine intermediate compound is transformed directly into the corresponding substitute 3-indole pyruvic acid by means of hydrolytic hydrogenation with sodium hyposulphate, in the presence of a Nickel-Raney catalyst in a buffered environment.

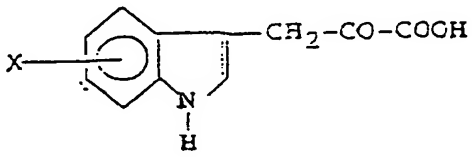
3. 6-chloro-tryptophan as an intermediate compound in the process according to claim 1.

4. 4-chloro-tryptophan as an intermediate compound in the process according to claim 1.

5. 5-fluoro-tryptophan as an intermediate compound in the process according to claim 1.

Patentansprüche

1. Verfahren zur Herstellung von in dem Benzolteil substituierten 3-Indol-Traubensaürederivaten, wie in der folgenden Formel dargestellt:



I

worin X Wasserstoff, Methyl, Methoxy oder Hydroxyl ist, in dem man von kernsubstituiertem Indol ausgeht, dadurch gekennzeichnet dass:
das substituierte Indol in einer basischen katalytischen Umgebung mit nieder Alkyl-3-(substituiertem Indol-3-yl)-2-Hydroxylimin-Propionat kondesiert wird und man die Oxylimin-Zwischenverbindung durch Reduktion mit wässrigem TiCl3 zum entsprechenden substituierten Tryptophan umwandelt, und
das substituierte Tryptophan in an sich bekannter Weise zur substituierten 3-Indol-Traubensäure umwandelt.

2. Verfahren gemäss Anspruch 1, worin die Oxyliminzwischenverbindung durch hydrolytische Hydrierung mit Natriumhyposulfat in Gegenwart eines Nickel-Raney Katalysators in einer Pufferumgebung unmittelbar in die entsprechende substituierte 3-Indoltraubensäure umgewandelt wird.

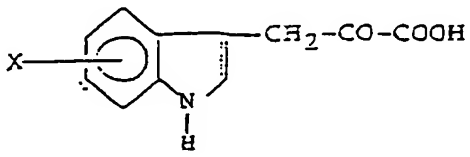
3. 6-Chlor-Tryptophan als Zwischenverbindung in dem Verfahren gemäss Anspruch 1.

4. 4-Chlor-Tryptophan als Zwischenverbindung in dem Verfahren gemäss Anspruch 1.

5. 5-Chlor-Tryptophan als Zwischenverbindung in dem Verfahren gemäss Anspruch 1.

Revendications

1. Procédé pour la production de dérivés de l' acide 3-indolpyruvique substitués dans leur moitié benzénique, représentés par la formule I



I

où X est hydrogène, halogène, méthyle, méthoxy ou hydroxyle, à partir d'un indole substitué dans son noyau, caractérisé en ce que:
le dit indole substitué est condensé, dans un environnement catalytique basique, avec une inf.alkylester-oxime de l' acide 3-bromo-pyruvique pour obtenir inf.alkyle-3-(indol-3-yle substitué)-2-hydroxylimine propionate; et
le dit composé oxylimine intermédiaire est transformé dans le correspondant tryptophane substitué par réduction avec TiCl3, et
le dit tryptophane substitué est transformé dans le dit acide 3-indolpyruvique de manière en soi connue.

2. Procédé selon la revendication 1, où le dit compose intermédiaire oxylimine est transformé directement dans son correspondent acide 3-indole-pyruvique au moyen d'une hydrogénation hydrolytique avec hyposulfate de sodium en présence d' un catalysateur Nickel-Raney dans un environnement tamponné.

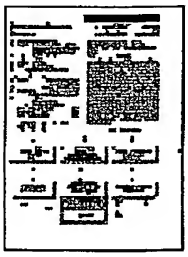
3. 6-chloro-tryptophane en tant que composé intermédiaire dans le procédé selon la revendication 1.

4. 4-chloro-tryptophane en tant que composé intermédiaire dans le procédé selon la revendication 1.

5. 5-chloro-tryptophane en tant que composé intermédiaire dans le procédé selon la revendication 1.

Title: **JP10245342A2: AGENT FOR REDUCING NEURAL CELL TOXICITY OF BETA-AMYLOID PROTEIN**

Derwent Title: Agent for reducing nerve cell toxicity of beta-amyloid protein - contains tea polyphenol [Derwent Record]



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1 pag

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Core: **C07D 311/00**; more...
IPC-7: **A61K 31/35**; A61K 35/78; C07D 311/62;

Priority Number: 1997-03-03 **JP1997000061761**

Abstract: PROBLEM TO BE SOLVED: To obtain an agent for reducing the neural cell toxicity of β-amyloid protein by using polyphenols contained in tea, especially tea catechins/ theaflavins, having storing activities for reducing the neural cell toxicity of the β-amyloid protein.

SOLUTION: The tea polyphenols used herein comprise tea catechins and theaflavins, and are used singly or in combinations. The tea catechins and theaflavins comprise a least one kind of epicatechin gallate, epigallocatechin gallate, thioflavin monogallate, theaflavin monogallate B and theaflavin digallate, and are obtained by extracting tea leaves with hot water, methanol, etc., dissolving the obtained extract solution in an organic solvent, evaporating off the organic solvent, and subsequently subjecting the residue to a high performance liquid chromatography. The agent for reducing the neural cell toxicity of the β-amyloid protein may be used singly or suitable together with auxiliary components in response to purposes.

Family: None

Other Abstract Info: DERABS C98-551152 DERC98-551152